5

10

15

20

25

## **CLAIMS**

1. A pair of probes for detecting and quantifying protein nuclear transport induced by action of a bioactive substance, comprising

Probe I in which a protein whose nuclear transport is to be detected or quantified is connected to an N-terminal end or a C-terminal end of a fusion protein [intein-C/reporter protein-C] wherein at least a C-terminal side polypeptide of an intein and a C-terminal side polypeptide of a reporter protein are connected in this order, and

Probe II in which a nuclear localization signal is connected to an N-terminal end or a C-terminal end of a fusion protein [reporter protein-N/intein-N] wherein at least the remaining N-terminal side polypeptide of the reporter protein and the remaining N-terminal side polypeptide of the intein are connected in this order.

2. A pair of probes for detecting and quantifying protein nuclear transport induced by action of a bioactive substance, comprising

Probe I in which a protein whose nuclear transport is to be detected or quantified is connected to an N-terminal end or a C-terminal end of a fusion protein [reporter protein-N/intein-N] wherein at least a N-terminal side polypeptide of a reporter protein and a N-terminal side polypeptide of an intein are connected in this order, and

Probe II in which a nuclear localization signal is connected to an N-terminal end or a C-terminal end of a fusion protein [intein-C/reporter protein-C] wherein at least the remaining C-terminal side polypeptide of the intein and the remaining C-terminal side polypeptide of the reporter protein are connected in this order.

3. The pair of probes of claim 1 or 2, wherein the intein is a DnaE intein derived from blue-green algae.

10

15

20

- 4. The pair of probes of claim 1 or 2, wherein the reporter protein is luciferase.
- 5 5. A method for detecting and quantifying protein nuclear transport induced by action of a bioactive substance, which comprises

making Probe I of the pair of probes of any of claims 1 to 4 and the bioactive substance coexist in the cytosol,

localizing Probe II in the nucleus, and measuring a signal of the reporter protein within the nucleus.

- 6. The detecting and quantifying method of claim 5, wherein polynucleotides expressing the pair of probes of any of claims 1 to 4 are introduced into a cell thereby making Probe I and the bioactive substance coexist in the cytosol and localizing Probe II in the nucleus.
- 7. The detecting and quantifying method of claim 5, wherein polynucleotides expressing the pair of probes of any of claims 1 to 4 are introduced into a non-human animal multipotent cell and the cell is subjected to ontogenesis thereby making Probe I and the bioactive substance coexist in the cytosol and localizing Probe II in the nucleus in all cells of the animal or its progeny.
- 8. A method for screening a protein nuclear transport-inducing substance, which comprises

introducing Probe I of the pair of probes of any of claims 1 to 4 into the cytosol,

localizing Probe II in the nucleus,

introducing a nuclear transport-inducing candidate substance into the cytosol, and



5

15

20

25

measuring a signal of the reporter protein in the nucleus.

9. A method for screening a protein nuclear transport-inhibiting substance, which comprises

introducing Probe I of the pair of probes of any of claims 1 to 4 into the cytosol,

localizing Probe II in the nucleus,

introducing a nuclear transport-inhibiting candidate substance into the cytosol,

further introducing a nuclear transport-inducing substance into the cytosol,

measuring a signal of the reporter protein in the nucleus, and comparing the signal with a signal of the reporter protein obtained by introducing only the protein nuclear transport-inducing substance into the cytosol.

- 10. The screening method of claim 8 or 9, wherein polynucleotides expressing the pair of probes of any of claims 1 to 4 are introduced into the cell thereby introducing Probe I into the cytosol and localizing Probe II in the nucleus.
- 11. The screening method of claim 8 or 9, wherein polynucleotides expressing the pair of probes of any of claims 1 to 4 are introduced into a non-human animal multipotent cell and the cell is subjected to ontogenesis thereby introducing Probe I in the cytosol and localizing Probe II in the nucleus in all cells of the animal or its progeny.